

Auxins are a class of plant growth substance (often called phytohormones or plant hormones) which plays an essential role in the coordination of many growth and behavioral processes in the plant life cycle and were the first class of growth regulators discovered. Indolacetic acid was the first auxin discovered, however, other chemicals also make up the auxin class of hormones. From the very first mitotic division of the zygote, gradients of auxin guide the patterning of the embryo into the parts that will become the organs of the plant: shoot apex, primary leaves, cotyledon(s), stem, and root. Depending on the specific tissue, auxin may promote axial elongation (as in shoots), lateral expansion (as in root swelling), or isodiametric expansion (as in fruit growth). The term 'Auxin' was firstly coined by F. W. Went.

The first plant hormone we will consider is auxin. Auxin deserves pride of place in any discussion of plant hormones because it was the first growth hormone to be discovered in plants, and much of the early physiological work on the mechanism of plant cell expansion was carried out in relation to auxin action.

Moreover, both auxin and cytokinin differ from the other plant hormones and signaling agents in one important respect: They are required for viability. Thus far, no mutants lacking either auxin or cytokinin have been found, suggesting that mutations that eliminate them are lethal. Whereas the other plant hormones seem to act as on/off switches that regulate specific developmental processes, auxin and cytokinin appear to be required at some level more or less continuously.

Indoacetic Acid

- a. Only naturally occurring auxin (IAA)
- b. Synthesized from the amino acid tryptophan
- c. A number of synthetic auxins have been produced

KEY POINTS

- Indoleacetic acid (IAA) was the first plant hormone discovered and is the primary auxin, but other substances also carry out the elongation function and are referred to as auxins.
- First detected in the coleoptiles of grass because auxin stimulated elongation of the grass coleoptiles
- Synthesized in the apical meristem of stems.
- Auxin moves only by polar transport (in one direction), that is, it moves down from the tip (where it is produced) through the stem. The rate of cell-to-cell transport of auxin is about 1 cm/hour.
- Polar transport does not depend on gravity (turning plants upside down results in the movement of auxin upward). It occurs because of a higher concentration of auxin transport proteins at the base of a cell, where they can transport the hormone to the apical end of the next cell. Auxin movement is based on uneven distribution of auxin efflux carriers on the plasma membrane, which send auxins in the proper direction. Pin-formed (PIN) proteins play a vital role in transporting auxin.
- Gradients of auxin guide the patterning of the embryo into the parts that will become the organs of the plant.
- Name of one Auxin binding receptor is ABP1.

Functions of Auxin:

The following are some of the responses that auxin is known to cause (Davies, 1995; Mauseth, 1991; Raven, 1992; Salisbury and Ross, 1992).

- Stimulates cell elongation.
 - (1) Does this by breaking the cross bonds of the cellulose polymers so that the cell can expand and then reforms them again
 - (2) The acid-growth hypothesis suggests that auxin causes hydrogen ions to be pumped into the cell wall, activating enzymes which break the bonds, making the wall flexible for expansion
 - (3) Turgor pressure causes the cell wall to expand
- Stimulates cell division in the cambium and, in combination with cytokinins in tissue culture
- Stimulates differentiation of phloem and xylem
- Stimulates root initiation on stem cuttings and lateral root development in tissue culture
- Mediates the tropistic response of bending in response to gravity and light (Geotropism and Phototropism).

Phototropism

- (1) Sunlight causes differential flow of auxin down the stem. More auxin flows down the dark side of the stem than the one on the light side.
- (2) Thus more elongation of the cells occurs on the dark side of the stem and causes the stem to bend toward the light
- (3) IAA (natural auxin only) is destroyed by sunlight]

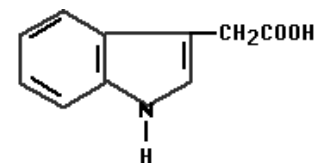
- The auxin supply from the apical bud suppresses growth of lateral buds i.e. shows Apical dominance

Apical dominance

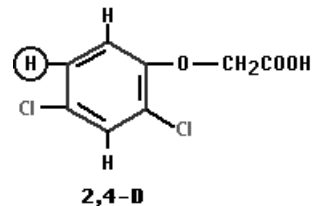
- (1) Auxin inhibits the development of lateral buds
 - (2) Does this by inhibiting the formation of a vascular connection between the lateral bud and the stem.
 - (3) Removing the apical bud removes the source of auxin and the lateral buds begin to grow]
- Delays leaf senescence
 - Can inhibit or promote (via ethylene stimulation) leaf and fruit abscission
 - Can induce fruit setting and growth in some plants
 - IAA applied to flowers can cause fruit development without fertilization (Parthenocarpy)
 - IAA produced by seeds in the fruit stimulates the pericarp to develop
 - Involved in assimilate movement toward auxin possibly by an effect on phloem transport
 - Delays fruit ripening
 - Promotes flowering in Bromeliads
 - Stimulates growth of flower parts
 - Promotes (via ethylene production) femaleness in dioecious flowers
 - Stimulates the production of ethylene at high concentrations

Synthetic auxins as weed killers

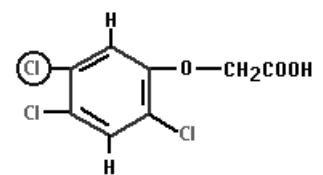
- Some of the most widely-used weed killers are synthetic auxins. These include 2,4-dichlorophenoxy acetic acid (2,4-D) and 2,4,5-trichlorophenoxy acetic acid (2,4,5-T).
- As the formulas show, 2,4,5-T is 2,4-D with a third chlorine atom, instead of a hydrogen atom, at the 5 position in the benzene ring (blue circles).
- 2,4-D and its many variants are popular because they are **selective herbicides**, killing broad-leaved plants but not grasses (no one knows the basis of this selectivity).
- Why should a synthetic auxin kill the plant? It turns out that the auxin influx transporter works fine for 2,4-D, but that 2,4-D cannot leave the cell through the **efflux transporters**. Perhaps it is the resulting accumulation of 2,4-D within the cell that kills it.
- A mixture of 2,4-D and 2,4,5-T was the "agent orange" used by the U.S. military to defoliate the forest in parts of South Vietnam.
- Because of health concerns, 2,4,5-T is no longer used in the U.S.
- NAA (naphthaleneacetic acid) is used to stimulate root development on cuttings for asexual propagation.



Indole-3-acetic acid (IAA)



2,4-D



2,4,5-T

Inhibitors of Auxin Transport Block Auxin Efflux

Several compounds have been synthesized that can act as **auxin transport inhibitors (ATIs)**, including NPA (1-Naphthylphthalamic acid) and TIBA (2,3,5-tri iodobenzoic acid) (Figure 19.16). These inhibitors block polar transport by preventing auxin efflux.

Brassinosteroids: Similar to Auxins

- Brassinosteroids are very similar to **auxins**, and for many years they were thought of as a type of **auxin**
- BRs have been shown to be involved in numerous plant processes: Works with **auxin** to promote cell expansion and cell elongation Has an unclear role in cell division and cell wall regeneration Promotes vascular differentiation

Promotes pollen elongation for pollen tube formation
 Accelerates the senescence in dying tissue cultured cells
 Provides some protection to plants during chilling and drought stress
 Although experimental evidence points to interactions of BR with other plant growth substances such as **auxin**, gibberellin, abscisic acid, ethylene, and jasmonate, the relationship of BR with these hormones has been documented primarily in plant growth regulatory processes

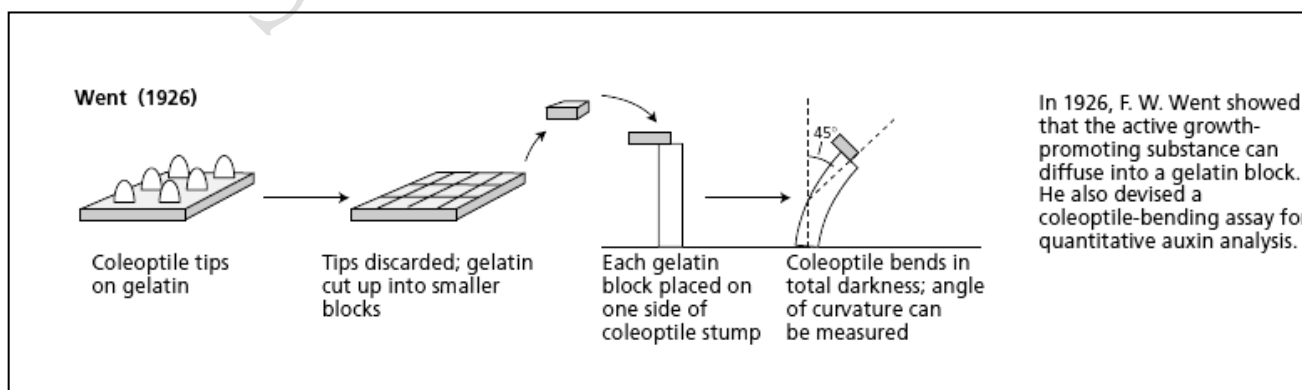
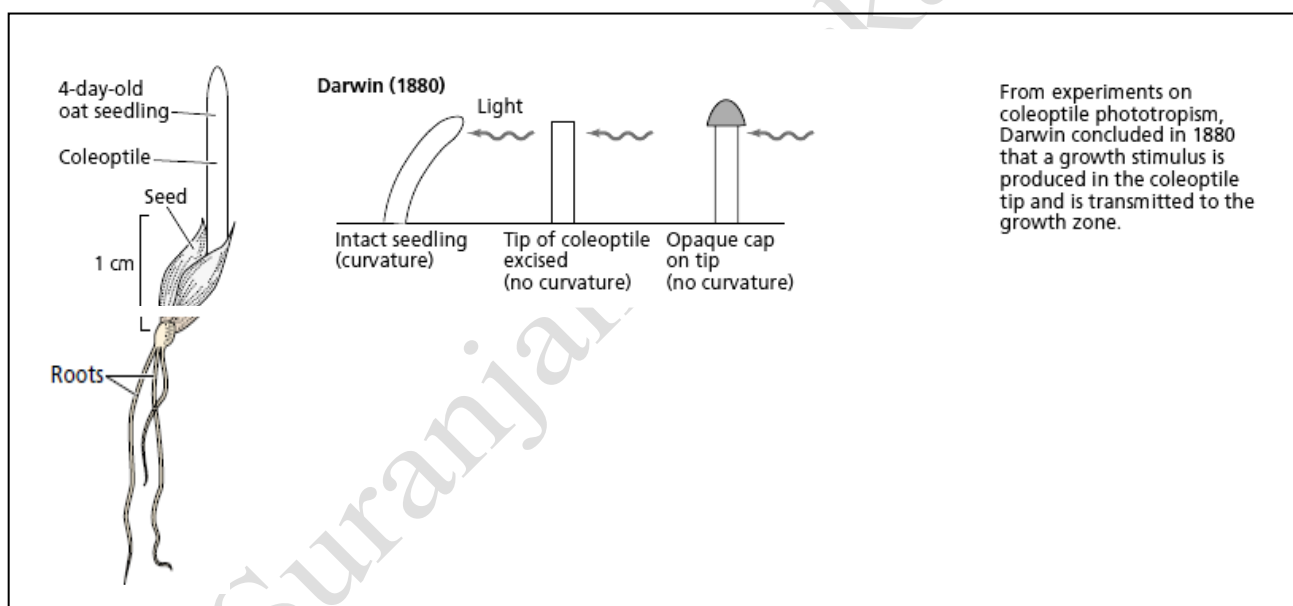
- Furthermore, with the exception of BR-**auxin** interaction, little is known in terms of how BR interacts with other hormones

Famous Experiments:

In 1926, a graduate student from Holland by the name of Fritz Went published a report describing how he isolated a plant growth substance by placing agar blocks under coleoptile tips for a period of time then removing them and placing them on decapitated Avena stems (Went, 1926). After placement of the agar, the stems resumed growth (see below). In 1928, Went developed a method of quantifying this plant growth substance. His results suggested that the curvatures of stems were proportional to the amount of growth substance in the agar (Went, 1928). This test was called the avena curvature test.

Darwin, C. (1880). *The Power of Movement in Plants*. (London: John Murray).

Went, F. (1935). Auxin, the plant growth hormone. *Bot. Rev.* **1**, 162–182.

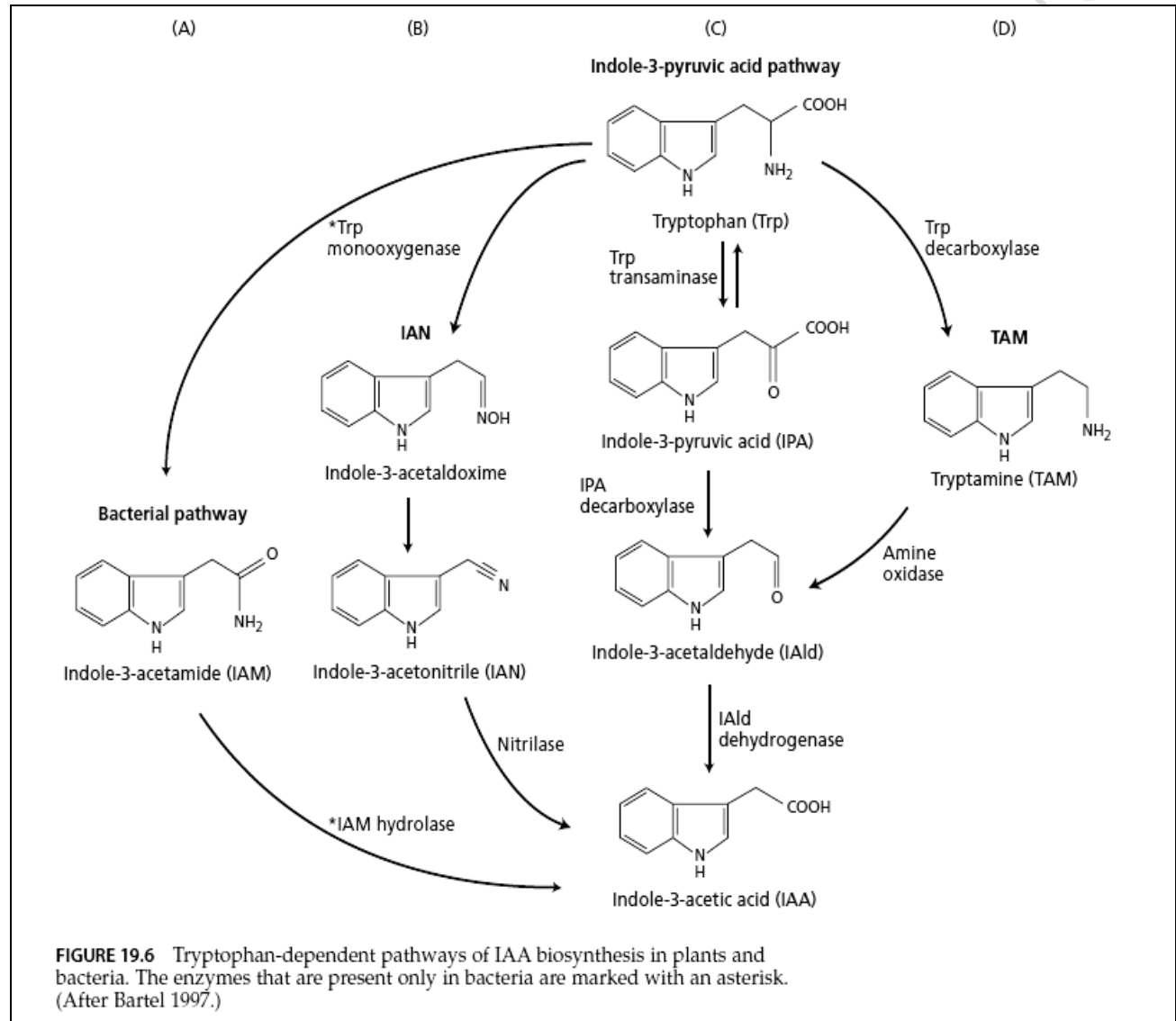


BIOSYNTHESIS OF AUXIN

Multiple Pathways Exist for the Biosynthesis of IAA

IAA is structurally related to the amino acid tryptophan, and early studies on auxin biosynthesis focused on tryptophan as the probable precursor. However, the incorporation of exogenous labeled tryptophan (e.g., [3H] tryptophan) into IAA by plant tissues has proved difficult to demonstrate. Nevertheless, an enormous body of evidence has now accumulated showing that plants convert tryptophan to IAA by several pathways, which are described in the paragraphs that follow.

The IPA pathway. The **indole-3-pyruvic acid (IPA)** pathway (see Figure 19.6C), is probably the most common of the tryptophan-dependent pathways. It involves a deamination reaction to form IPA, followed by a decarboxylation reaction to form indole-3-acetaldehyde (IAld). Indole-3-acetaldehyde is then oxidized to IAA by a specific dehydrogenase.



The TAM pathway. The **tryptamine (TAM) pathway** (see Figure 19.6D) is similar to the IPA pathway, except that the order of the deamination and decarboxylation reactions is reversed, and different enzymes are involved. Species that do not utilize the IPA pathway possess the TAM pathway. In at least one case (tomato), there is evidence for both the IPA and the TAM pathways (Nonhebel et al. 1993).

The IAN pathway. In the **indole-3-acetonitrile (IAN) pathway** (see Figure 19.6B), tryptophan is first converted to indole-3-acetaldoxime and then to indole-3-acetonitrile. The enzyme that converts IAN to IAA is called *nitrilase*. The IAN pathway may be important in only three plant families: the Brassicaceae (mustard family), Poaceae (grass family), and

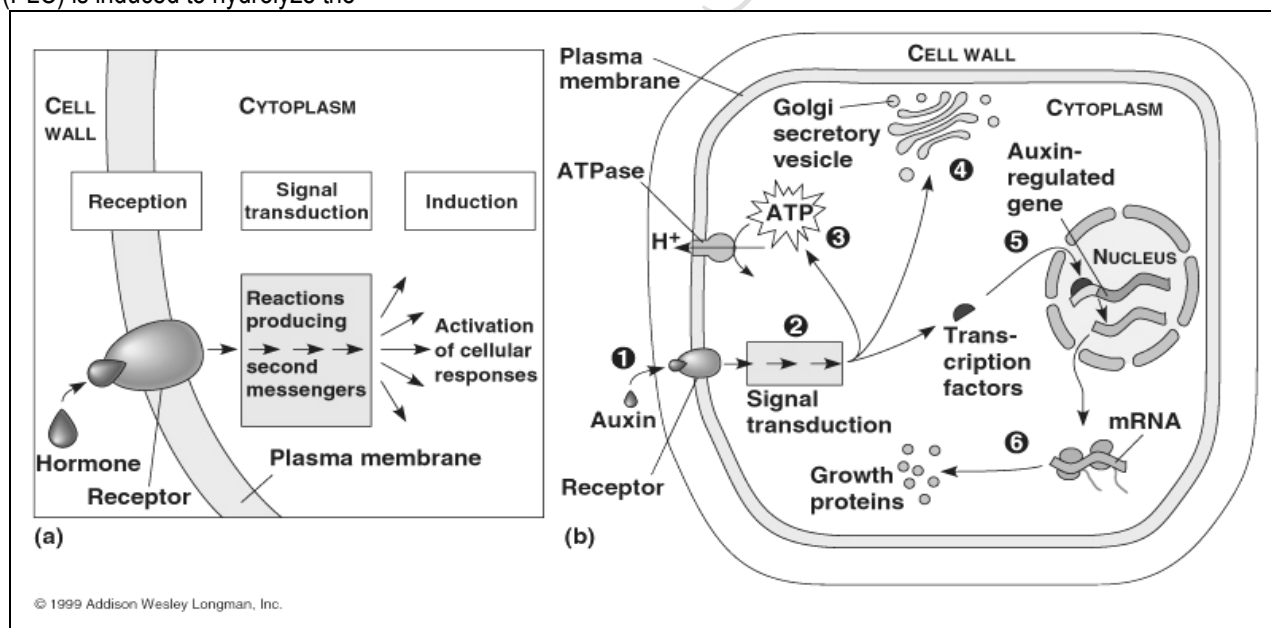
Musaceae (banana family). Nevertheless, nitrilase- like genes or activities have recently been identified in the Cucurbitaceae (squash family), Solanaceae (tobacco family), Fabaceae (legumes), and Rosaceae (rose family). Four genes (*NIT1* through *NIT4*) that encode nitrilase enzymes have now been cloned from *Arabidopsis*. When *NIT2* was expressed in transgenic tobacco, the resultant plants acquired the ability to respond to IAN as an auxin by hydrolyzing it to IAA (Schmidt et al. 1996). Another tryptophan-dependent biosynthetic pathway— one that uses **indole-**

3-acetamide (IAM) as an intermediate (see Figure 19.6A)—is used by various pathogenic bacteria, such as *Pseudomonas savastanoi* and *Agrobacterium tumefaciens*. This pathway involves the two enzymes tryptophan monooxygenase and IAM hydrolase. The auxins produced by these bacteria often elicit morphological changes in their plant hosts.

In addition to the tryptophan-dependent pathways, recent genetic studies have provided evidence that plants can synthesize IAA via one or more tryptophan-independent pathways. The existence of multiple pathways for IAA biosynthesis makes it nearly impossible for plants to run out of auxin and is probably a reflection of the essential role of this hormone in plant development.

Auxin and Signal Transduction:

The putative auxin receptors, which are responsible for transducing auxin signals into physiological changes, recognize auxin and other auxinic molecules (i.e. 2,4-D) because of certain structural characteristics. These receptors are then thought to signal a cascade of events leading to physiological responses or plant death in the case of auxinic herbicides. The mechanism for the signal-transduction pathway may involve direct interaction of regulatory proteins with specific DNA sequences or secondary messengers. Protein kinases, calcium-calmodulin, phosphoinositide metabolism, cyclic AMP, pH changes, redox reactions at the cell surface, membrane fatty acids and protein methylation have all been implicated in secondary messenger pathways in plants. For example, once auxin binds to a receptor, phospholipase C (PLC) is induced to hydrolyze the



membrane lipid phosphatidylinositol (PIP₂) which releases inositol-1,4,5-triphosphate (IP₃) and diacylglycerol (DAG). IP₃ and DAG can act as secondary messengers. The released IP₃ moves to the vacuolar membrane, the tonoplast, and binds to a receptor in that membrane. The IP₃ receptor then activates a calcium (Ca⁺⁺) transporter that pumps Ca⁺⁺ out of the vacuole, where it is stored, into the cytosol. Increased Ca⁺⁺ in the cytosol activates protein kinases that then activate other enzymes which catalyze the various reactions causing the metabolic changes associated with auxin or auxinic herbicide. Diacylglycerol (DAG) can also activate protein kinases that induce metabolic changes. The animation 'Auxin and Auxinic Herbicides Mechanisms of Action' illustrates this example of the signal transduction pathway.

[In growing shoots, auxin is transported unidirectionally, from the apex down the shoot. Along this pathway, the hormone enters a cell at the apical end, exits at the basal end, diffuses across the wall, and enters the apical end of the next cell. A pH difference between the cell wall (acidic at about pH 5) and the cytoplasm (pH 7) contributes to auxin transport. In the pH 7 environment of the cell, auxin is an anion. (1) When auxin encounters the acidic environment of the wall, the molecule picks up a hydrogen ion to become electrically neutral. (2) As a relatively small, neutral molecule, auxin passes across the plasma membrane. (3) Once inside a cell, the pH 7 environment causes auxin to ionize. This temporarily traps the hormone within the cell, because the plasma membrane is less permeable to ions than to neutral molecules of the same size. (4) ATP-driven proton pumps maintain the pH difference between the inside and outside of the cell. (5) Auxin can only exit the cell at the basal end, where specific carrier proteins are built into the membrane. The proton pumps contribute to this auxin efflux by generating a membrane potential (voltage) across the membrane, which favors transport of anions out of the cell. Now in the acidic environment of the wall again, auxin picks up a hydrogen ion and enters the next cell as an electrically neutral molecule. Polar auxin transport is one specific application of a basic mechanism of energy coupling in cells. This mechanism, chemiosmosis, uses proton pumps to store energy in the form of an H^+ gradient and membrane potential, and then taps this energy source to drive cellular work.]

Acid Growth Hypothesis and Cell Elongation Responses

A classic response to auxin or auxinic herbicides is cell elongation. Within 10 to 15 minutes of exposure to auxin, cell elongation is induced because of acid-induced cell wall loosening. The model best describing this process is the Acid Growth Hypothesis. Cell walls become acidified by IAA turning on H^+ -ATPases via secondary messengers, or by IAA inducing, via changes in gene expression, more H^+ -ATPase production for incorporation into the cell membrane.

The H^+ -ATPases pump H^+ out of the cell, expending ATP to move H^+ against its electrochemical gradient, further acidifying the cell-wall region. Wall loosening during acidification involves rearrangements of the load-bearing bonds of the cell wall and probably is mediated through special proteins called expansins. Once the cell wall is loosened, water moves into the cell, driven by higher internal osmotic pressure, thereby causing the cell to expand. Water uptake into the cells creates an internal turgor pressure that pushes against the cell membrane and extends the cell wall. Cell elongation after 30 to 60 minutes does not involve acid-induced elongation, but is due to auxin turning on genes which help cells elongate by other mechanisms (i.e. synthesis of new cell wall material). The animation 'Overall Picture of Auxinic Herbicide Action' illustrates how auxin induces the H^+ -ATPases pump H^+ out of the cell to cause cell wall loosening.

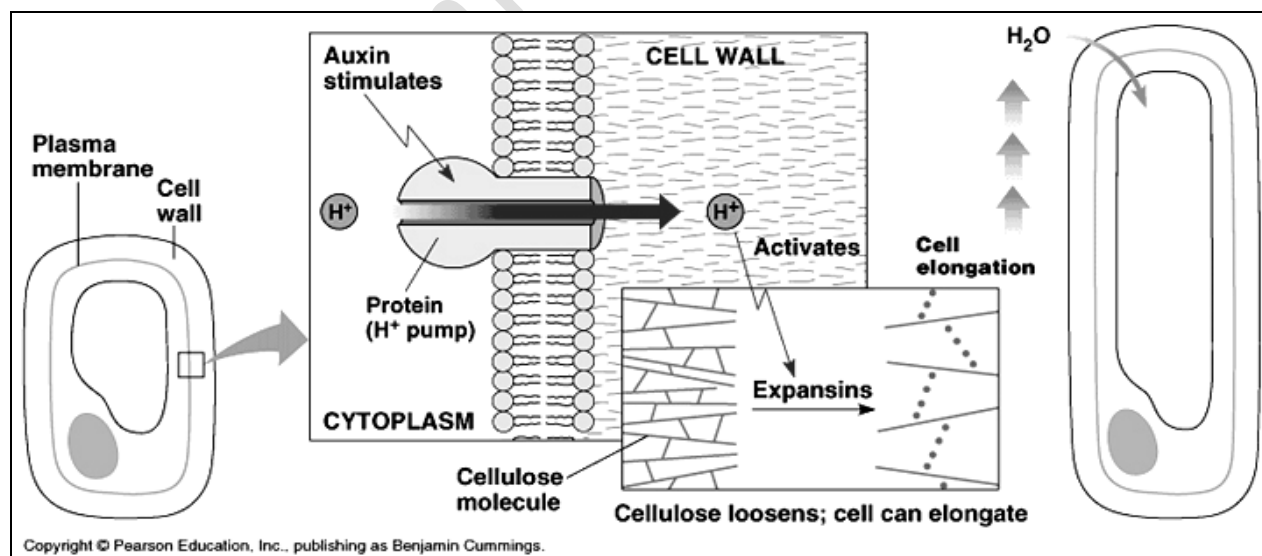


Figure : The chemiosmotic model for polar auxin transport (source: Taiz L., Zeiger E., 2010)